SUPPLEMENTAL FIGURE LEGENDS

Supplemental Figure S1: The -10 kb upstream enhancer up-regulates C/EBPα mediated activation of the PPARγ2 promoter. A fragment of the -10 kb region was amplified and inserted into a pGL3 (firefly luciferase) vector containing 1.6 kb of the upstream PPARγ2 promoter. Increasing amounts of a plasmid encoding C/EBPα were introduced at 0, 25, 50, 100, 200, and 400 ng. Activation of a plasmid containing renilla luciferase was used as a control. The value of relative luciferase units was set to the negative control of 0 ng of C/EBPα plasmid on the -1.6 kb control reporter. Results are from three independent samples performed in triplicate. Error bars: Mean + SD ($p^* < 0.05$, $p^{**} < 0.01$, $p^{***}<0.001$ by Student's two tailed *t*-test).

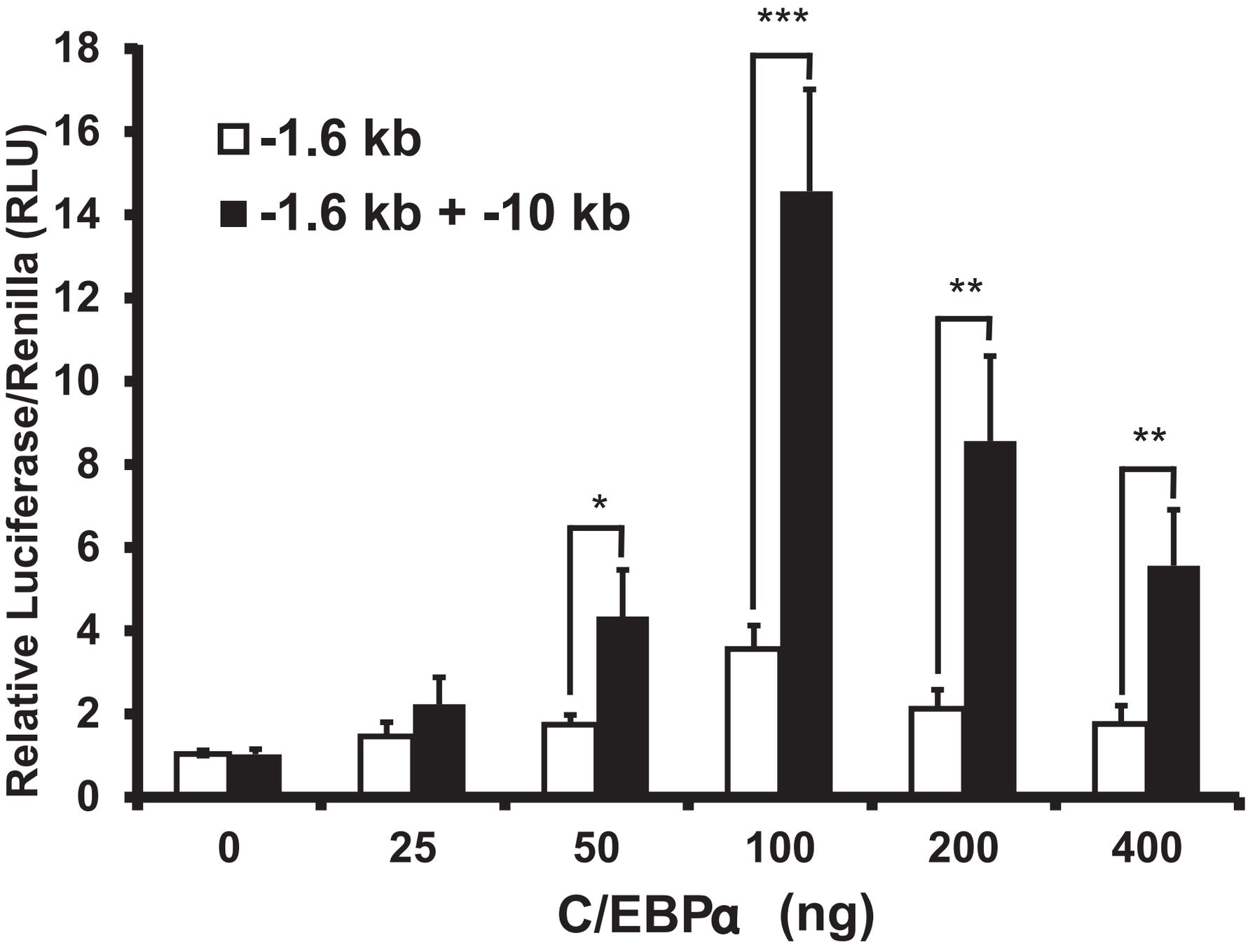
Supplemental Figure S2: Loop formation at the PPARγ2 locus in 3T3-L1 cells requires Prmt5. **(A)** Time course of interaction frequency between the promoter and the -10 kb enhancer sequence. 3T3-L1 cells were taken at the 0 hour timepoint and at 3, 6, 12, 24, and 96 hours following differentiation. **(B-D)** 3T3-L1 cells were infected with retroviruses containing either a control vector or antisense Prmt5 and differentiated. **(B)** Cells collected prior to differentiation (0 hour) or 96 hours later were tested by 3C for interactions between the PPARγ2 promoter and the -10 kb enhancer. Results are from three independent samples performed in triplicate. Error bars: Mean + SD ($p^* <0.05$, $p^{**} < 0.01$, by Student's two tailed *t*-test). **(C)** Oil Red O staining for cells differentiated for 4 days. **(D)** Western blots demonstrating a reduction in Prmt5 levels in antisense Prmt5 expressing cells. PI3K levels were monitored as a control.

Supplemental Figure S3: C/EBPα contributes to loop formation at the PPARγ2 locus. **(A)** Oil Red O staining of C3H10T/2 cells treated with a control siRNA or an siRNA poll targeting C/EBPα and differentiated for 4 days. **(B)** Western blot analysis of siRNA treated cells collected prior to differentiation (0 hour) or 96 hours later showing levels of C/EBPα. GAPDH levels were monitored as a control. **(C)** Cells collected prior to differentiation (0 hour) or 96 hours later were tested by 3C for interactions between the PPARγ2 promoter and the -10 kb enhancer. Results are from two independent samples performed in triplicate. Error bars: Mean + SD (p* <0.05, p** < 0.01, p***<0.001 by Student's two tailed *t*-test).

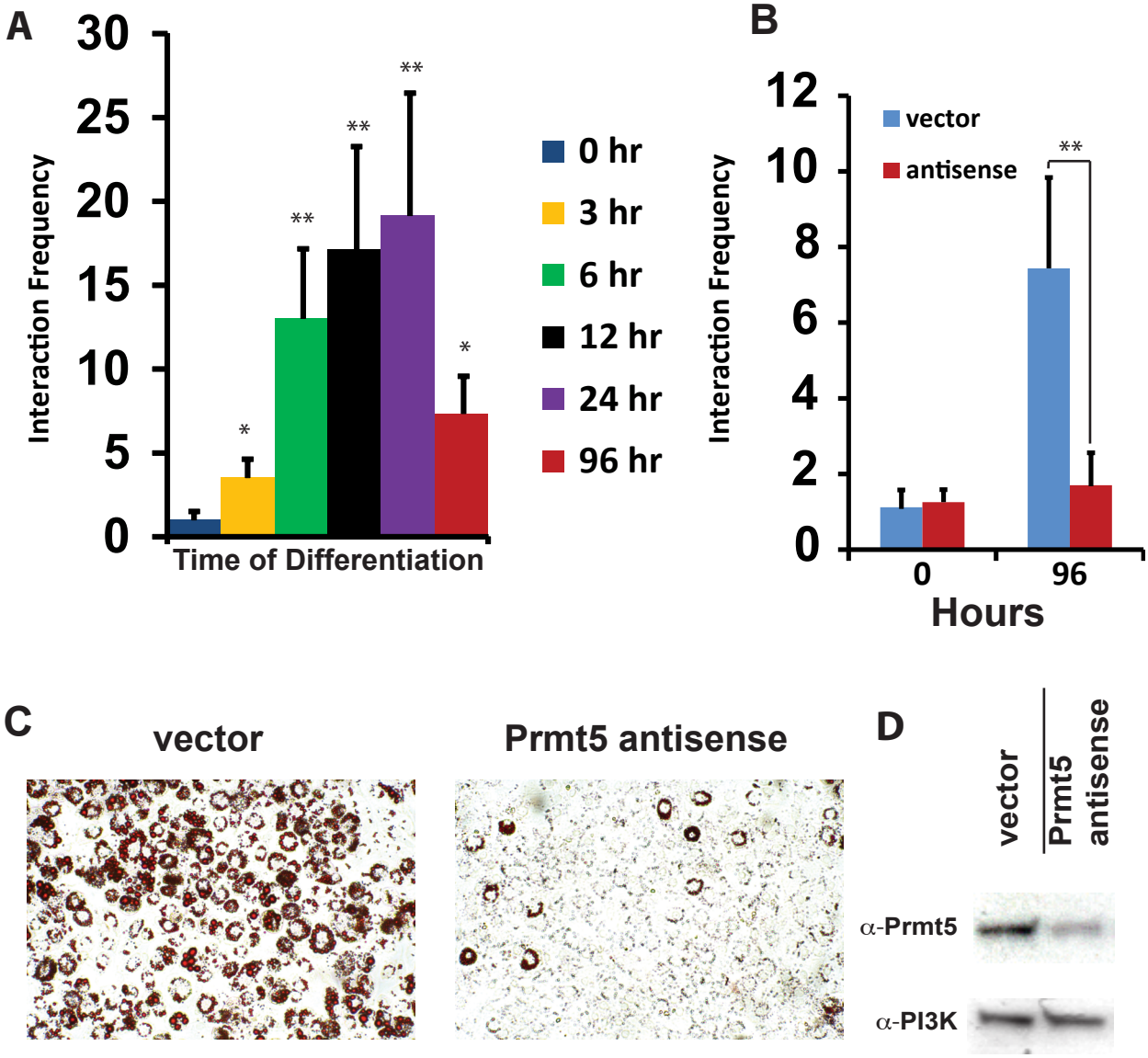
Supplemental Figure S4: Prmt5 mediates the formation of other DNA loops and inter-chromosomal interactions during adipogenic differentiation. **(A)** Time course of relative interaction frequency between the Ucp2 and Ucp3 loci. Samples of 3T3-L1 cells were taken at the 0 hour timepoint and at 3, 6, 12, 24, and 96 hours following differentiation. **(B)** Control vector and antisense Prmt5 expressing 3T3-L1 cells either collected prior to differentiation (0 hour) or 96 hours later were tested by 3C for interactions between the Ucp2 and Ucp3 loci. **(C)** Control vector and antisense Prmt5 expressing 3T3-L1 cells either collected prior to differentiation (0 hour) or 96 hours later were used to determine expression levels of Ucp3 and Ucp2, normalized to levels of GAPDH, by quantitative PCR. Data are presented as fold induction relative to the expression at time 0, which was set to 1. All results are from three independent samples performed in triplicate. Error bars: Mean + SD (p* <0.05, p** < 0.01, p***<0.001 by Student's two tailed *t*-test).

Supplemental Figure S5: Schematic model of the time course of events leading to DNA looping and enhanced transcription at the PPARγ2 locus. The progressively increasing thickness of the arrow at the PPARγ2 gene transcription start site represents increasing levels of productive transcription.

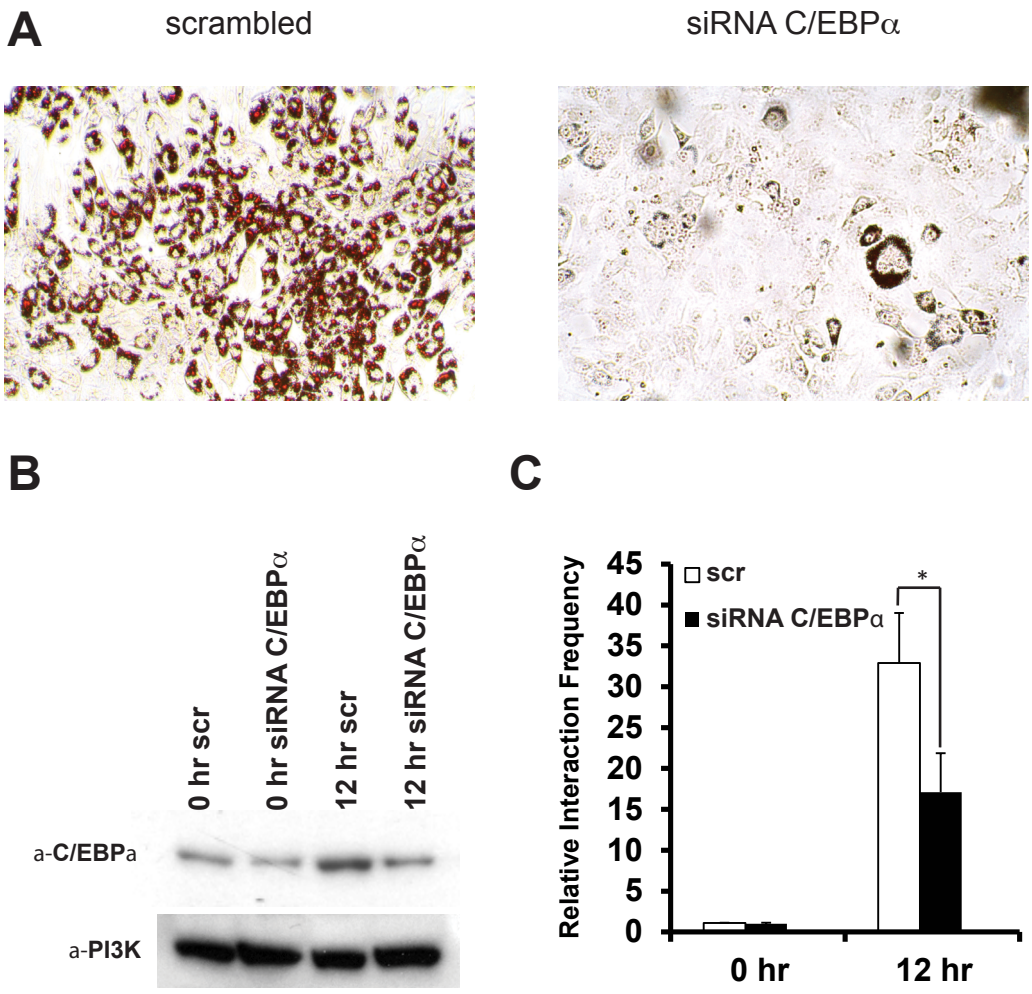
Supplemental Table 1: Primers used



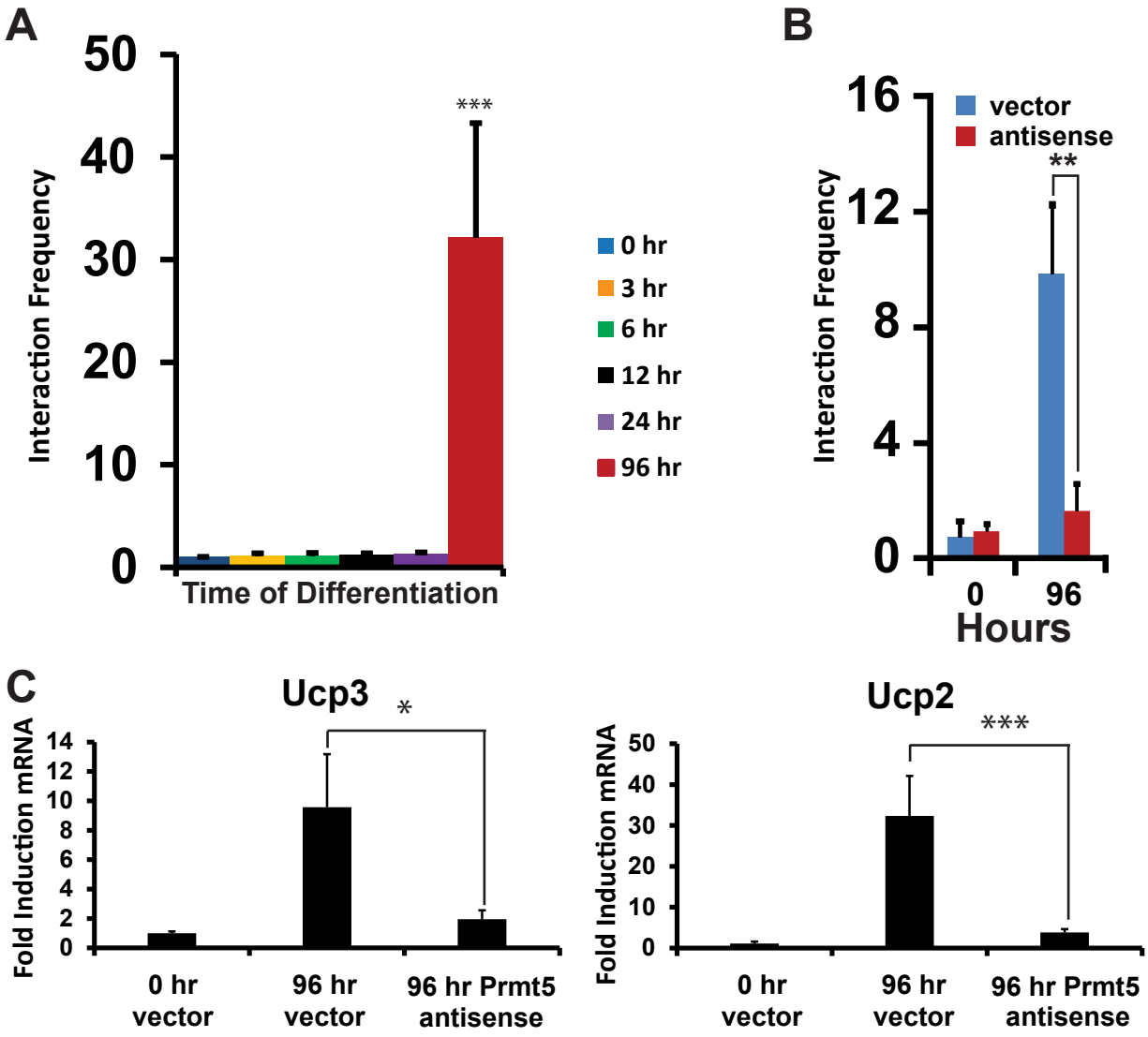
Supplemental Figure S1



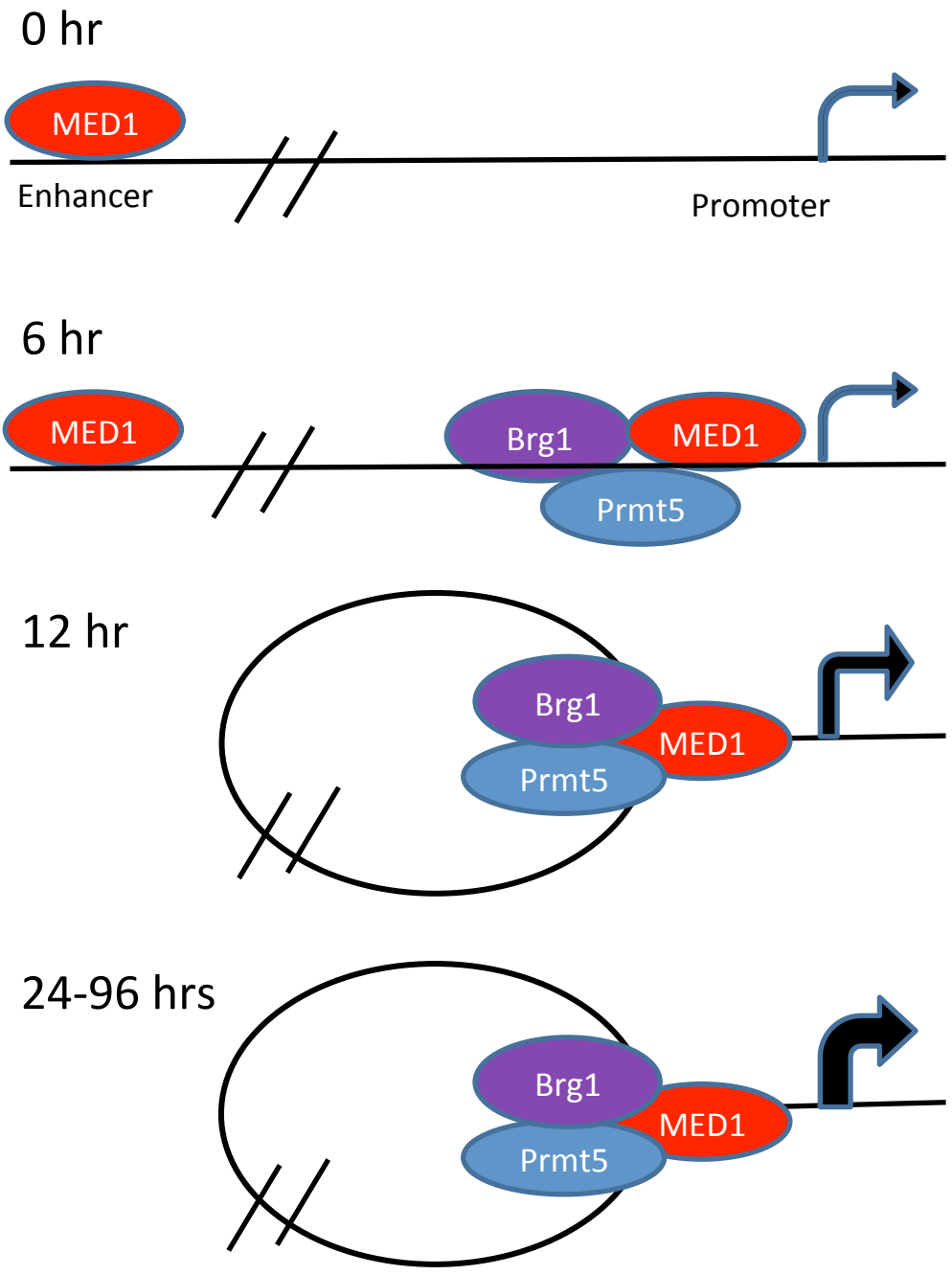
Supplemental Figure S2



Supplemental Figure 3C



Supplemental Figure S4



Supplemental Figure S5

Supplemental Table 1

Primers for Cloning

Forward	CGCGGGTACCGTTCCCAAAATGTTTATC
Reverse	CGCGGGTACCGTCTAGGAGGTAGGATATTCT

Primers for ChIP

PPARγ2 promoter	Forward	GTAATGTACCAAGTCTTGCCAAAGCA
	Reverse	AAGGAAAAAGGCTTATGGTCATCGAC
-10 kb region	Forward	TTCTTCCCAGTAGGAACTGCAT
	Reverse	GATCACTCAGTTGGCATTTCTC

Primers for 3C

Position (kb)	Restriction Enzyme	Sequence
PPARγ2		
-0.3 Anchor	**StuI**	**GTAATGTACCAAGTCTTGCCAAAGCA**
-81.6	PvuII	TGGTGTGGACAACCGATATG
-68	PvuII	AAGCTGAAGCCACACCCTAA
-54.6	PvuII	GAGGGCAATAGGGAAACACA
-38	StuI	AGGTGTGGCGTACACATCAG
-27	PvuII	TGTTGCTCTGGGCCTTTCTA
-20	PvuII	TCCAAGGCCTGACGCTATTA
-14.2	StuI	TGTAGCTAGGGGCAGTGTTG
-10	**PvuII**	**GGTGGACAAGCCAAAATCTTTA**
-5.5	StuI	GGACCGTAAGTCTGCCTGTTT
-3.7	PvuII	CACGTAAGTCGGGTACTCTCT
+2.1	PvuII	AGCCCATCAGCTTAGGCATT
+6.7	PvuII	TGGATTAGCTTCTTTCCCTAAGTG
+21.8	PvuII	TGCACCTTGGCAATTTAATG
+48	PvuII	GCCAGGTTATAGTTTGGTTTGG
+62	PvuII	TCAGGTCAGTGAGGGTAGGG
+71	StuI	GTGTAAGCCACAGGCAAGGT
+81	StuI	GTGTAAGCCACAGGCAAGGT
ERCC3		
-0.9	PvuII	GTCCACTGCTTTGAGCCTTC
13	StuI	AGCACTCTGGAAGCAGAAGC
Ucp3 +2	StuI	CTGTAGGGCAAAGGGGAACG
Ucp2 +0.6	PvuII	GACTGTATCCCCCAGGCACAGC

Expression Primers

PPARγ2	Forward	CCAGTGTGAATTACAGCAAATCTCTGTTTTAT
	Reverse	TTGTGAAGTGCTCATAGGCAGTG
Ucp3	Forward	AGACCCGATACATGAACGCT
	Reverse	TCCTGAGCCACCATCTTCAG
Ucp2	Forward	TTGTCTGGCAGCTGTGAAAC
	Reverse	GAAGCCATCACTTTAGGCCG
Cyclophilin	Forward	G GCCAACGATAAGAAGGG
	Reverse	GCTGTCTGTCTTGGTGCTCTCCAC

siRNA sequences

Prmt5 Oligo 3	Dharmacon D-042281-04	GUCCGUGCCUGUCGGGAAA
Prmt5 Oligo 5	(Invitrogen Stealth Prmt5 2075)	CCTACAGCACAGAAGGTGTAGAACA
MED1 Oligo 1	Ambion s72006	CACUGUCUCUCAUUGCAGtt
MED1 Oligo 2	Ambion s72008	CACUGCUAUUUUCUCAAUAtt